



GENOTYPE-SPECIFIC PHYSIOLOGICAL, BIOCHEMICAL, AND AGRONOMIC RESPONSES OF SOYBEAN TO WATER-DEFICIT STRESS

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Abstract

Water-deficit stress is one of the significant factors limiting soybean productivity, as it affects plant water relations, photosynthetic efficiency, biochemical metabolism, biomass production and reproductive development. The physiological, biochemical and agronomic response of soybeans to different water-deficit conditions was evaluated under different genotypes. Physiological characteristics, such as relative water content, chlorophyll content, leaf area index, and plant height biochemical characteristics, such as sugars and protein-related characteristics and agronomic characteristics, such as number of pods, biological weight, seed weight, seeds per pod, and seed yield, were evaluated in six soybean genotypes. There were significant genotype $\times$ stress interactions for plant height, relative water content, sugar concentration, biological weight and seed yield, showing that there are significant differences in the drought response among the genotypic groups. Stress, genotype and their interaction had significant effects on seed yield. Biological weight had the highest positive correlation with seed yield and was the most positive predictor in a regression model after removing the other traits. After adjusting for the other traits as their regression coefficients, chlorophyll A and sugar concentration were negatively correlated with seed yield, while biological weight had the highest positive coefficient. A combination of physiological, biochemical and agronomic variability was also reflected in drought response as shown by principal component analysis.

Keywords: Soybean; Water-deficit stress; Genotype $\times$ stress interaction; Physiological traits; Seed yield

1. Introduction

Soybean (*Glycine max* L.) is one of the most important leguminous crops worldwide because of its high-value protein and oil content and its major contribution to food, feed, and industrial production systems. However, soybean productivity is highly sensitive to environmental constraints, particularly water limitation during critical vegetative and reproductive growth stages. Drought can reduce canopy development, photosynthetic capacity, nutrient acquisition, biomass accumulation, reproductive success, and final seed yield. Developing soybean cultivars capable of maintaining productivity under limited water availability has therefore become an important breeding and crop-management objective, particularly under increasingly variable climatic conditions. Improving drought resilience requires a detailed understanding of the physiological, biochemical, and agronomic mechanisms that differentiate tolerant and susceptible soybean genotypes (Arya et al., 2021).

Soybean responses to drought are complex and involve coordinated changes in plant water relations, photosynthesis, stomatal behaviour, root development, metabolism, and stress-associated regulatory processes. Water deficit commonly decreases leaf water status, restricts cell expansion, alters gas exchange, and modifies photosynthetic pigment concentrations. Changes in root system architecture and transcriptional regulation also contribute to the ability of soybean plants to acquire water and sustain growth during drought conditions (Xiong et al., 2021). Experimental evidence further indicates that alterations in stomatal and hydraulic properties can improve water-use efficiency and reduce excessive water loss under drought, demonstrating the importance of physiological regulation in soybean adaptation to limited moisture availability (De Sousa et al., 2020). Similarly, substantial differences in plant growth, water consumption, and transpiration have been reported among soybean genotypes possessing contrasting drought-related characteristics (Minussi Winck et al., 2022).

Maintenance of leaf hydration is particularly important during water deficit because reductions in relative water content can affect membrane stability, cellular metabolism, photosynthesis, and plant growth. Genotypes capable of maintaining comparatively stable physiological function under drought may therefore possess stronger adaptive potential. Detailed physiological evaluations have demonstrated that drought tolerance in soybean involves coordinated regulation of water status, photosynthetic performance, antioxidant processes, and carbon metabolism rather than reliance on a single trait (Mesquita et al., 2020). Controlled studies have similarly shown that progressive water deficit substantially modifies soybean physiological characteristics, emphasizing the importance of simultaneously assessing multiple indicators when evaluating drought response (Wang et al., 2022). Genotype-dependent differences in physiological, biochemical, and genetic indicators further support the use of integrated trait-based approaches for identifying drought-tolerant soybean materials (Alzahrani, 2024).

Biochemical adjustment represents another central component of soybean drought adaptation. Water limitation can modify carbohydrate metabolism, protein synthesis, osmotic regulation, antioxidant activity, and stress-related gene expression. Accumulation or redistribution of soluble metabolites may contribute to osmotic adjustment and cellular protection during periods of declining water availability. Soybean plants capable of effective metabolic adjustment and subsequent recovery may exhibit improved resistance to severe water deficit and greater post-stress resilience (da Silva et al., 2022). The plant response can also be influenced by biological interactions within the rhizosphere; for example, drought tolerance may vary according to the associated *Bradyrhizobium* strain, indicating that plant–microbe interactions can modify physiological performance under water-limited conditions (Cerezini et al., 2020). Silicon-mediated improvements in vegetative growth and soil water retention have also demonstrated that drought response is closely connected with the capacity to preserve plant growth and water availability (Sah et al., 2022).

A major challenge in drought-tolerance research is the substantial variation observed among soybean genotypes. Different genotypes can exhibit contrasting responses in leaf water status, biomass accumulation, biochemical characteristics, nitrogen fixation, reproductive development, and yield. Genotypic variation in nitrogen fixation under drought has been demonstrated among short-season soybean cultivars, confirming that stress effects differ considerably according to genetic background (Liyanage et al., 2023). Genetic and agronomic screening has also identified drought-tolerant soybean genotypes and genomic regions associated with desirable agronomic characteristics, highlighting opportunities for breeding improvement (Chen et al., 2020). Multivariate drought screening based on multiple physiological and yield-related characteristics can therefore provide a more reliable classification of tolerant and susceptible genotypes than evaluation of individual traits alone (Yan et al., 2020).

The agronomic consequences of these physiological and biochemical responses are ultimately reflected in biomass production, pod formation, seed development, and yield. Physiological tolerance is agronomically valuable only when it contributes to maintenance of productivity under water limitation. Consequently, simultaneous evaluation of water status, photosynthetic characteristics, biochemical adjustment, growth, and yield is required to identify genotype-specific drought-response patterns and determine which traits are most closely associated with superior performance.

Objectives:

- □ Evaluate genotype-specific variation in physiological, biochemical, and agronomic traits of soybean under contrasting water-deficit conditions.
- □ Determine the effects of genotype, water stress, and genotype × stress interactions on key physiological and yield-related characteristics.
- □ Identify relationships among physiological, biochemical, and agronomic traits and determine the major characteristics associated with seed-yield performance under water-deficit stress.

2. Materials and Methods

2.1 Study Design and Data Source

This study was designed as a secondary data analysis to assess the physiological, biochemical and agronomic responses of soybean to water-deficit stress as a function of genotype. The analysis used data from the Advanced Soybean Agricultural Dataset (2025), which includes experimental observations from various soybean cultivars or hybrids under various treatment and water-stress conditions. The physiological, biochemical, growth, and yield-related traits are included in the dataset, which can be used for integrated assessment of drought response variability among soybean genotypes (Abdullah, 2025).

2.2 Experimental Factors and Dataset Structure

The data set consisted of six soybean genotypes (G1, G2, G3, G4, G5, and G6), three levels of water stress (S1, S2, and S3), two levels of treatment (C1 and C2), and three replicates of each experiment (R1, R2, and R3). Before analyzing the data, the original data set was checked for duplicate records and inconsistencies in the treatment code. 108 unique experimental observations were obtained by discarding exact duplicate observations. Treatment, stress level, genotype and replicate were separated from the composite treatment identifiers to allow factorial statistical analysis.

2.3 Physiological, Biochemical, and Agronomic Variables

Relative water content of leaves, chlorophyll A, B, leaf area index and plant height were used to evaluate physiological responses. Biochemical responses were estimated by sugar concentration, protein percentage and protein content. The agronomic performance was evaluated because of number of pods, biological weight, weight of 300 seeds, seed yield per unit area and number of seeds per pod. The variables were chosen to represent the different plant water status, photosynthetic capability, biochemical adjustment, vegetative growth and productivity changes under water deficit.

2.4 Data Preprocessing and Quality Control

Duplicate observations, missing values, inconsistent coding and implausible measurements were all checked in the dataset. To avoid pseudoreplication and overestimate of statistical significance, rows were removed to create exact copies. Summary statistics and graphical methods were used to analyze the continuous variables for unusual values and distributional patterns. Any potential outliers were kept in the data if there was no obvious data entry or measurement error. Any treatment label that was ambiguous in the preprocessing stage was not removed but flagged as such as it was not possible to clarify without documentation.

2.5 Descriptive Statistical Analysis

All the physiologic, biochemical and agronomic variables were subjected to descriptive statistics. For each trait, mean, standard deviation, minimum, maximum, and coefficient of variation were calculated. The distributions of traits were summarized across soybean genotype and water-stress level to detect general differences in soybean responses. Variation between the genotypes and between the treatments was shown graphically by boxplots and graphical comparison in groups.

2.6 Analysis of Genotype and Water-Stress Effects

A factorial analysis of variance was used in a general linear model (GLM) to assess the effects of soybean genotype and water-deficit stress. The principal explanatory factors included were genotype, water-stress level and their interaction. A statistically significant genotype $\times$ stress interaction was considered to be evidence of a genotype differential response to a water deficit. Additional treatment factor, as needed, was added to the model to represent the effects in isolation and interaction.

2.7 Post-Hoc Mean Comparison

If there were large main or interaction effects, means were compared with Tukey's honestly significant difference test. The procedure was utilized to determine the soybean genotype differences that were significant at each water-stress condition at a controlled family-wise error rate. Where interaction effects were present, the marginal means were estimated and then treated separately.

2.8 Correlation and Multivariate Analysis

Relationships among physiological, biochemical and agronomic traits were assessed using Pearson correlation analysis to evaluate associations between plant water status, chlorophyll characteristics, biochemical responses and seed yield. Standardized variables were subjected to principal component analysis to determine the major dimensions of variation and the traits that are primarily responsible for the differentiation of genotypic variation. To help classify soybean genotypes based on similarity in the drought-response profile, hierarchical cluster analysis and heatmap visualization were also performed.

2.9 Regression Analysis

The physiological and biochemical traits most closely related to seed yield per unit area were selected using multiple linear regression analysis. Relative water content, leaf area index, plant height, biological weight, pod number, sugar content and protein-related traits were used as possible predictors of seed yield. The assumptions of linearity, homoscedasticity, and normality were checked by examining residual diagnostics, while multicollinearity was checked by

using variance inflation factors.

2.10 Statistical Criteria and Software

All statistical tests were conducted at two-tailed levels of significance of $p < 0.05$. Prior to the use of inferential analysis, the assumptions of normality and homogeneity of variance were assessed, and alternative appropriate robust or non-parametric methods were considered if these assumptions were significantly violated. The collected data was analyzed and processed with IBM SPSS Statistics and Python, where the proper Python libraries were applied for data preprocessing, statistical modelling, multivariate data analysis, and graphical data visualization.

2.11 Ethical Considerations

The study did not include any human subjects or animals as experimental objects and was solely based on the Advanced Soybean Agricultural Dataset (2025) available to the public. For this reason, this study did not require institutional ethical approval and informed consent. The dataset was only used for academic research, and all the pre-processing steps were carried out without intentional manipulation of the original experimental data. Duplicate records have been eliminated solely for the purpose of improving analytical validity, whereas treatment labels were uncertain and kept in place with the records clearly marked as such to preserve the integrity of the source data.

3. Results

3.1 Overall Variation in Physiological, Biochemical, and Agronomic Traits

Significant differences were noted in the soybean observations of physiological, biochemical and agronomic traits. The mean height of plants was 49.96, and the mean number of pods was 144.94 as presented in Table 1. The biological weight had significant variation, from 32.00 to 236.00, and a mean of 114.53. The relative water content of leaves (RWCL) was 0.674 ± 0.059 , and varied between 0.434 and 0.875. The average chlorophyll A and B were 3.94 and 2.40 respectively. There was significant variability in seed yield per unit area, with a mean value of 4,388.41 and range 2,127.50 to 7,841.50. The relatively wide ranges were found in the biological weight, chlorophyll concentration, sugar content and seed yield data, which showed that there was a significant genotypic variation among the genotype and treatment combinations.

Table 1. Overall descriptive statistics of physiological, biochemical, and agronomic traits

Trait	Mean	SD	Minimum	Maximum
Plant Height (PH)	49.959	3.066	40.400	55.700
Number of Pods (NP)	144.936	20.114	75.000	218.000
Biological Weight (BW)	114.528	49.150	32.000	236.000
Sugars (Su)	0.478	0.237	0.161	1.115
Relative Water Content in Leaves (RWCL)	0.674	0.085	0.434	0.875
Chlorophyll A663	3.938	2.860	1.000	10.200
Chlorophyll b649	2.399	1.193	1.100	7.800
Protein Percentage (PPE)	36.043	2.428	31.100	39.900
Weight of 300 Seeds (W3S)	35.349	5.023	26.400	53.900
Leaf Area Index (LAI)	0.072	0.022	0.020	0.100
Seed Yield per Unit Area (SYUA)	4388.415	1257.257	2127.500	7841.500
Number of Seeds per Pod (NSP)	2.020	0.247	1.400	2.600
Protein Content (PCO)	0.508	0.289	0.120	1.800

3.2 Genotype-Specific Trait Differences

Significant variations were observed between the 6 soybean genotypes. The results in Table 2 indicate that Genotype 6 had the highest mean RWCL (0.709), followed by Genotypes 5 (0.702) and 1 (0.701). Mean levels of chlorophyll A (Genotype 3, 5.644) and biological weight (Genotype 3, 155.833) were highest, while mean number of pods was highest (Genotype 4, 156.094). Genotype 5 had the largest overall seed production per unit area (5,016.49), followed by Genotype 3 (4,682.92) and Genotype 1 (4,575.23). The lowest overall seed yield (3,655.12) was recorded by genotype 6, which had comparatively high leaf relative water content. These findings show that plant water status maintenance was not always associated with better agronomic productivity.

Table 2. Mean physiological, biochemical, and agronomic responses across soybean genotypes

Genotype	RWCL	Chlorophyll A	Sugars	Pods	Biological Weight	Seed Yield
G1	0.701	3.528	0.494	141.906	82.278	4575.233
G2	0.664	3.911	0.428	143.267	123.778	4441.567
G3	0.628	5.644	0.590	146.817	155.833	4682.922
G4	0.638	4.556	0.430	156.094	114.944	3959.161
G5	0.702	2.194	0.531	139.194	132.444	5016.489
G6	0.709	3.794	0.392	142.339	77.889	3655.117

When stress levels were considered simultaneously, the genotype-specific responses to the yield became more apparent. In this case, Genotype 3 recorded a significantly higher mean yield of 6,392.50 with S3 compared to S1 (3,586.52), and Genotype 5 had a mean yield of 6,138.18 under S3. In contrast, Genotype 4 decreased from 4,930.70 under S1 to 3,275.57 under S3. The percentage of genotype 6 was relatively low for all three stress categories. The different stress responses observed suggest a considerable genotype $\times$ stress interaction for yield. Figure 1 shows the Genotype-specific variation in mean seed yield per unit area across water-stress levels S1–S3.

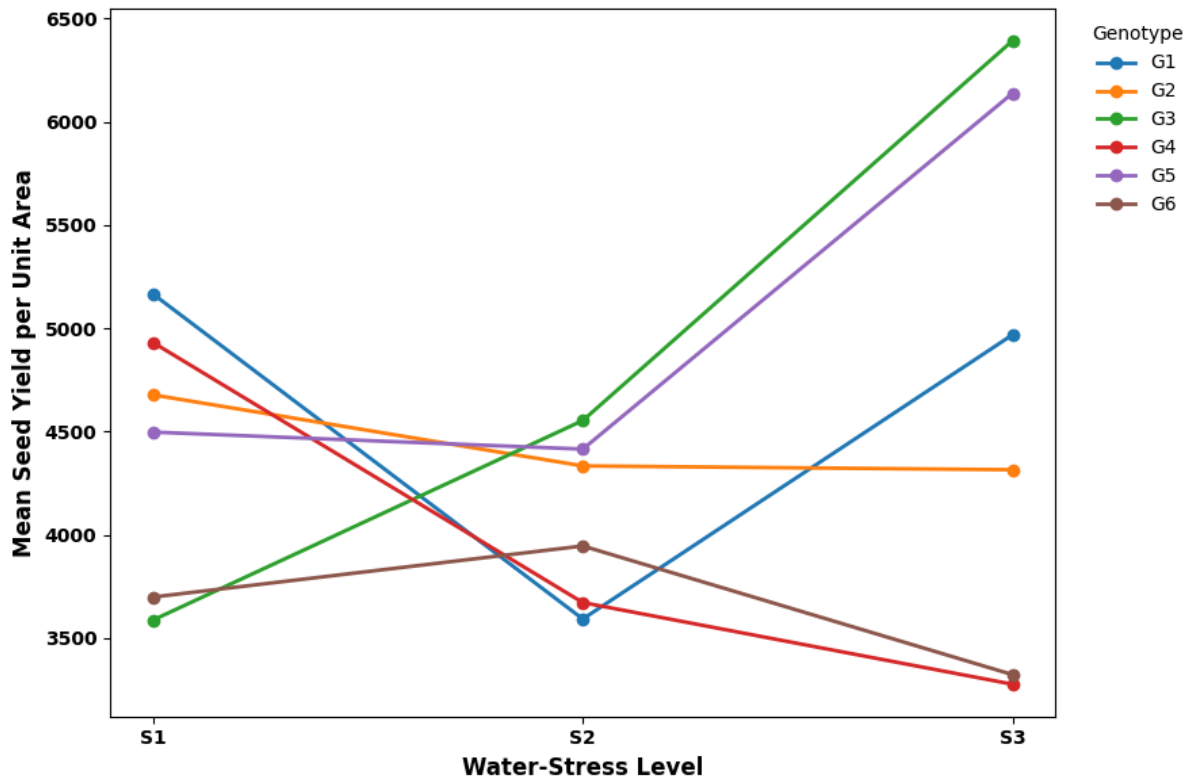


Figure 1. Genotype-specific variation in mean seed yield per unit area across water-stress levels S1–S3

3.3 Response Patterns across Water-Stress Levels

The mean responses of the water-stress coding are reported in Table 3. The stress categories were only slightly different with RWCL varying from 0.661 for S3 to 0.683 for S2. The chlorophyll A increased from 2.833 under S1 to 4.827 under S2 and was relatively high in S3 as 4.319. The sugar concentration decreased from 0.564 (S1) to 0.541 (S2) to 0.326 (S3). Mean biological weight was lowest under S2 (101.152) while mean seed yield was also lowest under S2 (4,042.23). S3 (4,735.19) had the highest overall mean seed yield. Due to the nature of the differences indicated, the categories of stress were not interpreted as being linear with an increase in severity from S1 to S3, but rather as being stress specific.

Table 3. Mean trait values according to coded water-stress level

Stress Level	RWCL	Chlorophyll A	Sugars	Pods	Biological Weight	Seed Yield
S1	0.677	2.833	0.564	143.736	120.795	4361.236
S2	0.683	4.827	0.541	145.152	101.152	4042.230
S3	0.661	4.319	0.326	146.039	120.000	4735.194

The differences in RWCL between individual genotypes were significantly higher than the differences between the overall averages of the stress levels. Genotype 3 showed high variability among the stress categories with mean values of RWCL of 0.613, 0.853 and 0.540 under S1, S2 and S3, respectively, as shown in Figure 2. Genotypes 5 and 6 had relatively high RWCL values under S3 (0.749 and 0.741), respectively. This heterogeneity also contributes to genotype-dependent physiological adaptation to water-stress conditions under experimental conditions.

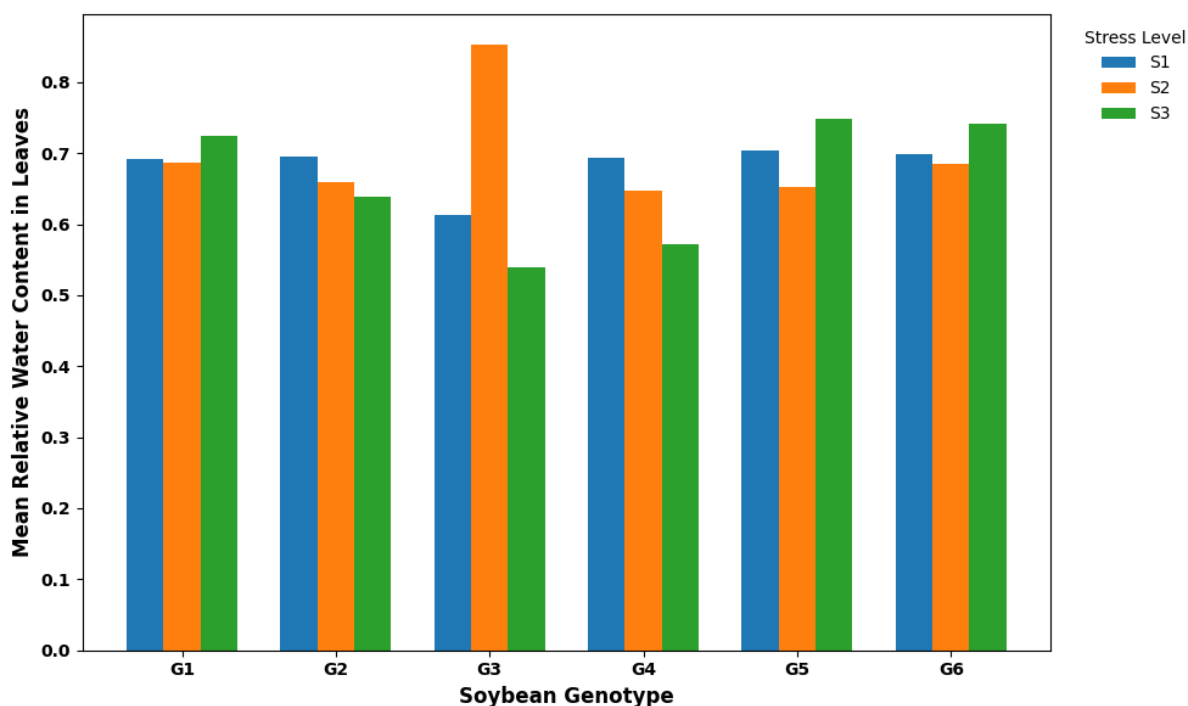


Figure 2. Mean relative water content in leaves of six soybean genotypes across water-stress levels S1–S3

3.4 Effects of Genotype, Water Stress, and Their Interaction

Several traits were found to be more strongly affected by the genotype-specific responses than by the main effect of stress alone by a factorial analysis. As shown in Table 4, plant height varied significantly among the genotypes ($F = 11.67$, $p < 0.001$), genotype $\times$ stress interaction was also significant ($F = 10.65$, $p < 0.001$), but stress effect was not significant.

This trend was seen at RWCL as well. The main stress effect was not significant ($F = 1.14$, $p = 0.324$), but the genotype differences were significant ($F = 5.59$, $p < 0.001$) as well as the stress $\times$ genotype interaction ($F = 6.90$, $p < 0.001$). The interaction between stress and genotype was not significant ($p = 0.075$), but stress ($F = 8.18$, $p < 0.001$) and genotype ($F = 4.64$, $p < 0.001$) had significant effects on chlorophyll A.

The sugar content was significantly associated with stress, genotype and the interaction of both (all $p \leq 0.004$). Biological weight was affected by stress ($F = 3.24$, $p = 0.044$), genotype ($F = 15.84$, $p < 0.001$), and genotype $\times$ stress interaction ($F = 4.97$, $p < 0.001$). Most importantly, seed yield exhibited significant effects of stress ($F = 6.10$, $p = 0.003$), genotype ($F = 6.05$, $p < 0.001$), and genotype $\times$ stress interaction ($F = 6.77$, $p < 0.001$). The number of pods produced did not have any statistically significant main or interaction effects.

Table 4. Factorial analysis of genotype and water-stress effects on selected soybean traits

Trait	Effect	F	p-value
Plant Height	Stress	1.13	0.329
	Genotype	11.67	<0.001
	Genotype $\times$ Stress	10.65	<0.001
RWCL	Stress	1.14	0.324
	Genotype	5.59	<0.001
	Genotype $\times$ Stress	6.90	<0.001
Chlorophyll A	Stress	8.18	<0.001
	Genotype	4.64	<0.001

	Genotype $\times$ Stress	1.79	0.075
Chlorophyll B	Stress	19.09	<0.001
	Genotype	2.56	0.033
	Genotype $\times$ Stress	3.95	<0.001
Sugars	Stress	23.69	<0.001
	Genotype	3.73	0.004
	Genotype $\times$ Stress	7.37	<0.001
Number of Pods	Stress	0.11	0.897
	Genotype	1.83	0.116
	Genotype $\times$ Stress	1.41	0.188
Biological Weight	Stress	3.24	0.044
	Genotype	15.84	<0.001
	Genotype $\times$ Stress	4.97	<0.001
Seed Yield	Stress	6.10	0.003
	Genotype	6.05	<0.001
	Genotype $\times$ Stress	6.77	<0.001

3.5 Relationships among Physiological, Biochemical, and Agronomic Traits

Several significant relationships were detected using a correlation analysis among characteristics measured. Biological weight had the highest positive correlation with seed yield ($r = 0.574$, $p < 0.001$) as presented in Table 5. There was also a positive correlation between the number of pods and the seed yield ($r = 0.217$, $p = 0.024$). However, chlorophyll A ($r = -0.244$, $p = 0.011$) and sugar concentration ($r = -0.255$, $p = 0.008$) showed significant negative correlation with yield. There was a weak and negative correlation between RWCL and yield ($r = -0.141$; $p = 0.146$).

Table 5. Pearson correlation matrix for selected physiological, biochemical, and agronomic traits

Trait	RWCL	Chl A	Sugars	Pods	Biological Weight	Seed Yield
RWCL	1.000	0.109	0.028	-0.170	-0.153	-0.141
Chlorophyll A	0.109	1.000	0.077	0.082	-0.064	-0.244*
Sugars	0.028	0.077	1.000	-0.040	0.076	-0.255**
Number of Pods	-0.170	0.082	-0.040	1.000	0.231	0.217*
Biological Weight	-0.153	-0.064	0.076	0.231	1.000	0.574***
Seed Yield	-0.141	-0.244*	-0.255**	0.217*	0.574***	1.000

*Note: * $p < 0.05$; ** $p < 0.01$; *** $p < 0.001$.

The correlation structure is illustrated in Figure 3. The comparatively high positive linkage between biological weight and seed yield is highlighted in the heatmap, while the rest of the physiological and biochemical traits were generally weak to moderate in their relatedness with seed yield. The overall biomass accumulation was a better indicator for the yield performance than any individual leaf-water or biochemical indicator showed.

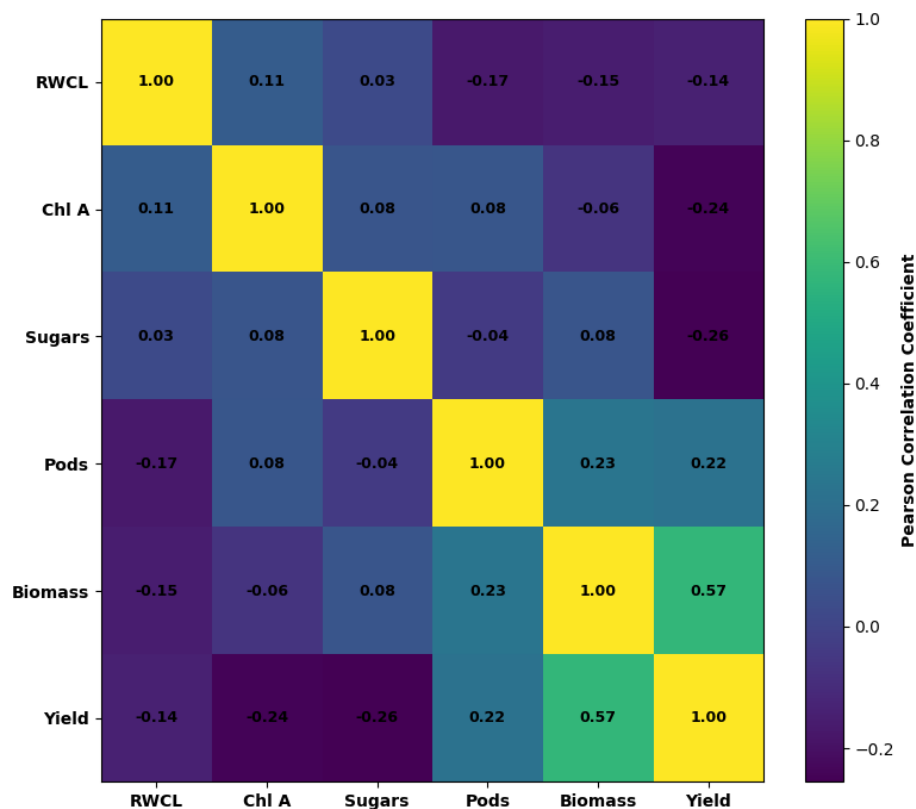


Figure 3. Correlation heatmap showing relationships among selected physiological, biochemical, and agronomic traits of soybean

3.6 Multivariate Differentiation of Soybean Responses

The overall variation of the entire set of physiological, biochemical and agronomic parameters was analysed by principal component analysis. The first principal component accounted for 17.32% and the second component accounted for 16.51% of the total standardized variance, which together accounted for 33.83% of variance.

The highest positive loading was observed for seed yield (0.559), weight of 300 seeds (0.341), number of seeds per pod (0.333) and plant height (0.318). The weight of sugar was negatively loaded on PC1 (-0.381). PC2 was primarily correlated with leaf area index (0.407), protein content (0.395) and biological weight (0.378), while chlorophyll B was negatively correlated (-0.369) and RWCL (-0.303).

Figure 4 shows that there was significant multivariate heterogeneity both within and between the different genotypes in the distribution of observations along PC1 and PC2. The moderate amount of variance accounted for by the first two components suggests that the drought-response differentiation was multidimensional and was not controlled by a single physiological or agronomic trait.

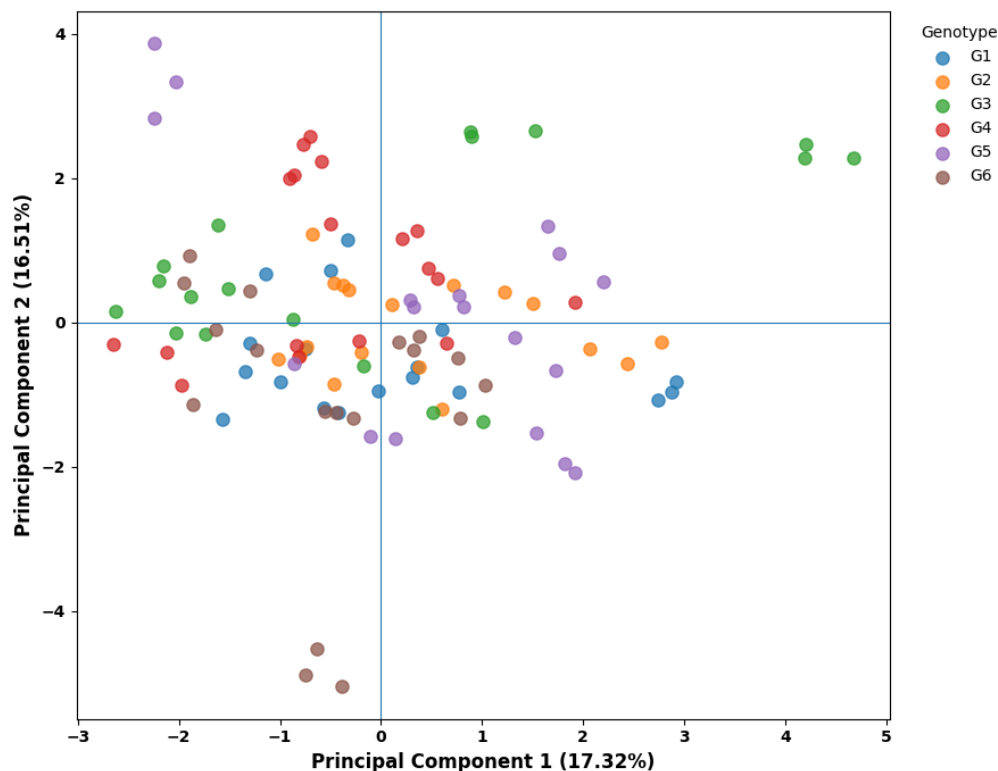


Figure 4. Principal component analysis of physiological, biochemical, and agronomic characteristics across soybean genotypes

3.7 Predictors of Seed Yield

Multiple regression analysis was then performed to determine the characteristics that are independent of seed yield. The overall model was statistically significant ($F = 10.36$, $p < 0.001$) and explained 51.6% of the variance in seed yield ($R^2 = 0.516$; adjusted $R^2 = 0.467$).

The strongest positive association with seed yield, as shown in Table 6. (standardized $\beta = 0.533$, $p < 0.001$), was with biological weight. Plant height also contributed positively ($\beta = 0.229$, $p = 0.003$). However, chlorophyll A ($\beta = -0.241$, $p = 0.003$) and sugar concentration ($\beta = -0.251$, $p = 0.004$) were significant negative predictors following simultaneous adjustment for the other variables. In the multivariable model, none of the following variables were independently associated with seed yield: RWCL, chlorophyll B, leaf area index, number of pods, protein percentage or protein content.

Table 6. Multiple regression analysis of physiological and biochemical predictors of seed yield

Predictor	Standardized β	t	p-value
RWCL	0.014	0.180	0.857
Chlorophyll A	-0.241	-3.102	0.003
Chlorophyll B	-0.114	-1.389	0.168
Leaf Area Index	0.039	0.512	0.610
Plant Height	0.229	3.070	0.003
Biological Weight	0.533	7.010	<0.001
Number of Pods	0.111	1.464	0.146
Sugars	-0.251	-2.956	0.004

Protein Percentage	0.053	0.704	0.483
Protein Content	−0.036	−0.442	0.659

In general, the findings show that the soybean reaction to the coded water-deficit treatments were highly genotype dependent. The genotype $\times$ stress interactions for plant height, RWCL, sugar concentration, biological weight and seed yield were significant, implying that the six genotypes had very different physiological adjustment and agronomic performance. Biological weight proved to be the strongest positive association and independent predictor of seed yield, and the multivariate analysis showed that the soybean stress response was not one single drought-response trait, but a composite of several of the physiological, biochemical and productivity-related traits measured.

4. Discussion

Soybean exhibited significant genotype differences in the physiology, biochemical and agronomic responses under the two water deficit treatments. Significant G $\times$ S interactions were found for plant height, RWC in the leaf, sugar concentration, biological weight, and seed yield, showing that these six soybean genotypes did not respond consistently to variations in water availability. This discovery aligns with prior studies that have demonstrated a high heritability of drought tolerance in soybean and that reflect the physiological adjustment, allocation of resources, and reproductive stability of different soybean genotypes under drought conditions (Soliman et al., 2025).

While there was no significant difference between stress levels, relative water content was very different between genotypes and had a significant genotype $\times$ stress interaction. This indicates that maintenance of tissue hydration was more related to genetic background than to the stress treatment alone. There were genotypes with relatively high leaf water content throughout the stress levels and genotypes with significant variation in leaf water content. The water status and physiological recovery under drought conditions were found to be genotype-dependent, with similar differences observed in soybean (Kulundžić et al., 2022). The diverse reaction seen in the present study could also be due to physiological memory or adaptive adjustment to water limitation, since soybean plants can be stress-induced in their physiology, which can affect their reactions to water limitation in later stages (Bester et al., 2024).

Water-stress conditions and genotype effects were significant for chlorophyll A and B. Changes in the water status of plants may affect various aspects of chloroplast structure, pigment biosynthesis and photosynthetic capacity, making chlorophyll pigments particularly vulnerable to plant water status changes. Water stress has been seen to significantly alter the chlorophyll concentration, canopy properties and vegetation indices between soybean cultivars in previous studies (Tavares et al., 2022). The results of the present study confirm that the chlorophyll properties can be useful indicators of the stress response, but they can differ between genotypes and may not necessarily reflect final productivity. Stress, genotype and their interaction had significant effects on the sugar concentration. This highlights the significance of carbohydrate metabolism in drought osmotic adjustment and energy redistribution. Soluble sugars can be stored as compatible osmolytes, help maintain cellular structure and protect against dehydration. This negative correlation between sugar levels and yield in the current analysis, however, implies that higher sugar accumulation also may reflect higher physiological stress, and not necessarily higher productivity. A similar metabolic reorganization during drought is reported in soybean where alterations in sugar, antioxidants, and other metabolites were strongly correlated with drought severity (Saraswat et al., 2020). These biochemical responses are also affected by beneficial microbial associations which further affect plant performance and increase tolerance to drought stress by improving nutrient uptake and stress protection (Sheteiwy et al., 2021).

Biological weight was one of the most significant agronomic parameters in the present study. It varied greatly between stress levels and was significantly different among the different genotypes, and there was also a significant genotype $\times$ stress interaction. Most important, biological weight was the greatest positive correlate of yield and the most positive predictor in the multiple regression model. This suggests that there is a close relationship between the ability to sustain biomass production under water limitation and the reproductive productivity. This has been noted in soybean cultivars under drought stress during flowering and early seed development where maintaining growth and biomass were correlated with increased resilience and yield stability (Poudel et al., 2023).

Stress, genotype and genotype $\times$ stress interaction significantly influenced seed yield, thus revealing the importance of genotype-specific adaptation to water deficits. Some genotypes remained unaffected or showed improvement in yield in specific stress categories, other genotypes exhibited definite yield losses. This variation has also been shown through drought tolerance indices and evaluation based on heterosis, with soybean genotypes showing significant differences in their capacity to maintain productivity under water-limited conditions (Mejaya et al., 2025). The present results, therefore, further emphasize the need to consider G $\times$ E interactions, instead of considering mean performance alone.

The strong negative correlation of chlorophyll A and sugar concentration with seed yield imply that more vigorous physiological or biochemical reactions may not necessary result in increased productivity. Drought tolerance, however, seems to rely on concerted regulation of several traits. The multidimensional nature of soybean adaptation is further supported by responses in metabolism, anatomy, and physiology when the plants are subjected to combined environmental stress. Responses of soybeans to combined environmental stress in terms of metabolism, anatomy, and physiology add to the multidimensional nature of adaptation. Likewise, emerging multi-omics evidence suggests that drought resistance is not just a collection of traits, but rather the result of a coordinated regulation of root physiology, metabolism and stress-responsive molecular pathways (Wang et al., 2024). These integrated responses are also known to

be controlled by hormones, especially as drought stress intensifies (Zhou et al., 2022).

In general, results suggest that the development of drought tolerance in soybean should be viewed as an integrated genotype-dependent process that includes water relations, photosynthesis, biochemical adjustments, biomass maintenance and yield stability.

5. Conclusion

The present study showed that the soybean adaptation to water-deficit stress involves coordinated physiological, biochemical and agronomic adjustments which are highly genotype dependent. Significant genotype $\times$ stress interactions indicated that there were significant differences among the genotypes in their responses to varying water availability for plant height, relative water content, sugar concentration, biological weight, and yield of seed. The relationships between relative water content and chlorophyll parameters with productivity were not consistently positive, suggesting that there is no single physiological parameter that explains drought tolerance. Biological weight proved to be the most significant agronomic characteristic affecting productivity among traits measured. It had the highest positive correlation with seed yield, and it was the most significant positive predictor in the multivariable regression model. The other significant positive effect on yield was plant height, while sugar concentration and chlorophyll A had significant negative relationships, after controlling for the other variables. The results indicate that maintenance of biomass accumulation and overall growth stability might be more strongly correlated with productive drought adaptation rather than individual biochemical/pigment responses. In general, the findings are in favor of the trait-based approach to an integrated system for selection of soybean cultivars with better water-deficit adaptation. The genotypic variability observed serves as a valuable reference for identifying good material for drought-tolerance breeding and should be considered further research in controlled field-based drought intensities and environments.

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